CLAIMS

1. An isolated peptide fragment of the conserved regulatory domain of NFAT protein capable of inhibiting protein-protein interaction between calcineurin and NFAT, or a biologically active analog thereof.

2. The peptide fragment or biologically active analog thereof of claim 1, further being capable of inhibiting dephosphorylation of said NFAT by said calcineurin.

3. The peptide fragment or biologically active analog thereof of claim 2, further being capable of inhibiting a conformational change in said NFAT that results from said dephosphorylation of said NFAT by said calcineurin.

4. The peptide fragment or biologically active analog thereof of claim 1, further being capable of inhibiting recruitment of said NFAT to the nucleus in a cell.

5. The peptide fragment or biologically active analog thereof of claim 1, further being capable of inhibiting a conformational change in said NFAT that results from said protein-protein interaction between said NFAT and said calcineurin.

6. The peptide fragment or biologically active analog thereof of claim 1, further being capable of inhibiting NFAT-dependent gene transcription.

7. The peptide fragment or biologically active analog thereof of claim 1 wherein said fragment or biologically active analog thereof does not inhibit the activity of calcineurin toward non-NFAT calcineurin substrates.

8. The peptide fragment of claim 1 wherein said conserved regulatory domain of NFAT protein is selected from the group consisting of NFAT1, NFAT2, NFAT3 and NFAT4.

9. The peptide fragment of claim 1 wherein said conserved regulatory domain of NFAT protein is human or murine.

10. The peptide fragment of claim 1 wherein said interaction between said calcineurin and said NFAT is between said calcineurin and a member selected from the group consisting of NFAT1, NFAT2, NFAT3 and NFAT4.

11. The peptide fragment of claim 1 wherein said fragment is less than about 150 amino acid residues in length.

12. The peptide fragment of claim 1 wherein said fragment is less than about 100 amino acid residues in length.

13. The peptide fragment of claim 1 wherein said fragment is less than about 50 amino acid residues in length.

14. The peptide fragment of claim 1 wherein said fragment is less than about 30 amino acid residues in length.

15. The peptide fragment of claim 1 wherein said fragment is less than about 20 amino acid residues in length.

16. The peptide fragment of claim 1 wherein said fragment is less than about 10 amino acid residues in length.

17. The peptide fragment of claim 1 wherein said fragment is less than about 6 amino acid residues in length.

18. The peptide fragment of claim 1 wherein said fragment is greater than about 3 amino acid residues in length.

19. The peptide fragment of claim 1 wherein said fragment comprises the amino acid sequence IX_2X_3T (SEQ ID NO:104) and wherein X_2 is E, R or Q, and X_3 is I or F.

20. The peptide fragment of claim 19 wherein said fragment comprises the amino acid sequence IEIT (SEQ ID NO:105).

21. The peptide fragment of claim 19 wherein said fragment comprises the amino acid sequence IRIT (SEQ ID NO:106).

22. The peptide fragment of claim 19 wherein said fragment comprises the amino acid sequence IQIT (SEQ ID NO:107).

23. The peptide fragment of claim 19 wherein said fragment comprises the amino acid sequence IQFT (SEQ ID NO:108).

24. The peptide fragment of claim 1 wherein said fragment comprises the amino acid sequence $X_1IX_2X_3T$ (SEQ ID NO:73) and wherein X_1 is R or S, X_2 is E, R or Q, and X_3 is I or F.

25. The peptide fragment of claim 24 wherein said fragment comprises the amino acid sequence X_1IX_2IT (SEQ ID NO:74).

26. The peptide fragment of claim 25 wherein said fragment comprises the amino acid sequence RIX_2IT (SEQ ID NO:75).

27. The peptide fragment of claim 25 wherein said fragment comprises the amino acid sequence X_1IEIT (SEQ ID NO:76).

28. The peptide fragment of claim 24 wherein said fragment comprises the amino acid sequence RIEIT (SEQ ID NO:1).

29. The peptide fragment of claim 24 wherein said fragment comprises the amino acid sequence SIRIT (SEQ ID NO:2).

30. The peptide fragment of claim 24 wherein said fragment comprises the amino acid sequence SIQIT (SEQ ID NO:3).

31. The peptide fragment of claim 24 wherein said fragment comprises the amino acid sequence SIQFT (SEQ ID NO:4).

32. The peptide fragment of claim 24 wherein said fragment comprises the amino acid sequence $PX_1IX_2X_3T$ (SEQ ID NO:77).

33. The peptide fragment of claim 32 wherein said fragment comprises the amino acid sequence selected from the group consisting of:

PRIEIT (SEQ ID NO:5),

PSIRIT (SEQ ID NO:6),

PSIQIT (SEQ ID NO:71), and

PSIQFT (SEQ ID NO:7).

34. The peptide fragment of claim 32 wherein said fragment comprises the amino acid sequence $X_5PX_1IX_2X_3T$ (SEQ ID NO:78), and wherein X_5 is S or C.

35. The peptide fragment of claim 32 wherein said fragment comprises the amino acid sequence selected from the group consisting of:

SPRIEIT (SEQ ID NO:8),

CPSIRIT (SEQ ID NO:9),

CPSIQIT (SEQ ID NO:10), and

CPSIQFT (SEQ ID NO:11).

36. The peptide fragment of claim 34 wherein said fragment comprises the amino acid sequence $X_5PX_1IX_2X_3TX_6$ (SEQ ID NO:79), and wherein X_6 is P or S.

37. The peptide fragment of claim 36 wherein said fragment comprises the amino acid sequence selected from the group consisting of:

SPRIEITP (SEQ ID NO:12),

SPRIEITS (SEQ ID NO:13),

CPSIRITS (SEQ ID NO:14),

CPSIQITS (SEQ ID NO:15), and

CPSIQFTS (SEQ ID NO:16).

38. The peptide fragment of claim 36 wherein said fragment comprises the amino acid sequence $X_5PX_1IX_2X_3TX_6X_7$ (SEQ ID NO:80), and wherein said X_7 is S, C or I.

39. The peptide fragment of claim 38 wherein said fragment comprises the amino acid sequence selected from the group consisting of:

SPRIEITPS (SEQ ID NO:17),

SPRIEITSC (SEQ ID NO:18),

CPSIRITSI (SEQ ID NO:19),

CPSIQITSI (SEQ ID NO:20), and

CPSIQFTSI (SEQ ID NO:21).

40. The peptide fragment of claim 38 wherein said fragment comprises the amino acid sequence $X_{11}X_{10}X_9X_5PX_1IX_2X_3TX_6X_7X_8$ (SEQ ID NO:81), and wherein X_8 is H, L or S; X_9 is P, L or E; X_{10} is G, L or F; and X_{11} is S, A, V or P.

41. The peptide fragment of claim 40 wherein said fragment comprises the amino acid sequence selected from the group consisting of:

SGPSPRIEITPSH (SEQ ID NO:22),

SGLSPRIEITPSH (SEQ ID NO:23),

ALESPRIEITSCL (SEQ ID NO:24),

VLECPSIRITSIS (SEQ ID NO:25),

PFECPSIQITSIS (SEQ ID NO:26),

PFECPSIQITSIS (SEQ ID NO:27), and

PFECPSIQFTSIS (SEQ ID NO:28).

42. The peptide fragment of claim 40 wherein said fragment comprises the amino acid sequence selected from the group consisting of:

KPAGASGPSPRIEITPSHELMQAGG (SEQ ID NO:29),

KPAGASGLSPRIEITPSHELIQAVG (SEQ ID NO:30),

PDGAPALESPRIEITSCLGLYHNNN (SEQ ID NO:31),

AGGGRVLECPSIRITSISPTPEPPA (SEQ ID NO:32),

LGGPKPFECPSIQITSISPNCHQEL (SEQ ID NO:33),

LGGPKPFECPSIQITSISPNCHQGT (SEQ ID NO:34), and

LGGPKPFECPSIQFTSISPNCQQEL (SEQ ID NO:35).

43. An isolated polynucleotide comprising a polynucleotide encoding the peptide comprising the amino acid sequence as set forth in SEQ ID NO:104, and biologically active analogs thereof.

44. An isolated polynucleotide comprising a member selected from the group consisting of:

a polynucleotide encoding the peptide comprising the amino acid sequence as set forth in SEQ ID NO:105, SEQ ID NO:106, SEQ ID NO:107, SEQ ID NO:108, and biologically active analogs thereof.

45. The isolated polynucleotide of claim 44 wherein said polynucleotide comprises a member selected from the group consisting of the nucleotide sequence as set forth in SEQ ID

NO:110, SEQ ID NO:111, SEQ ID NO:112, SEQ ID NO:113, SEQ ID NO:114 and SEQ ID NO:115.

46. The isolated polynucleotide of claim 44 wherein said polynucleotide is capable of hybridizing to and which is at least about 70% identical to the nucleotide sequence as set forth in SEQ ID NO:110, SEQ ID NO:111, SEQ ID NO:112, SEQ ID NO:113, SEQ ID NO:114 or SEQ ID NO:115.

47. An isolated polynucleotide comprising a member selected from the group consisting of:

a polynucleotide encoding the peptide comprising the amino acid sequence as set forth in SEQ ID NO:73, SEQ ID NO:74, SEQ ID NO:75, SEQ ID NO:76, and biologically active analogs thereof.

48. An isolated polynucleotide comprising a member selected from the group consisting of:

a polynucleotide encoding the peptide comprising the amino acid sequence as set forth in SEQ ID NO:1, SEQ ID NO:2, SEQ ID NO:3, SEQ ID NO:4, and biologically active analogs thereof.

49. The isolated polynucleotide of claim 48 wherein said polynucleotide comprises a member selected from the group consisting of the nucleotide sequence as set forth in SEQ ID NO:36, SEQ ID NO:37, SEQ ID NO:38, SEQ ID NO:39, SEQ ID NO:83 and SEQ ID NO:84.

50. The isolated polynucleotide of claim 48 wherein said polynucleotide is capable of hybridizing to and which is at least

about 70% identical to the nucleotide sequence as set forth in SEQ ID NO:36, SEQ ID NO:37, SEQ ID NO:38, SEQ ID NO:39, SEQ ID NO:83 or SEQ ID NO:84.

51. An isolated polynucleotide comprising a member selected from the group consisting of:

a polynucleotide encoding the peptide comprising the amino acid sequence as set forth in SEQ ID NO:77, SEQ ID NO:78, SEQ ID NO:79, SEQ ID NO:80, SEQ ID NO:81, and biologically active analogs thereof.

52. An isolated polynucleotide comprising a member selected from the group consisting of:

a polynucleotide encoding the peptide comprising the amino acid sequence as set forth in SEQ ID NO:5, SEQ ID NO:6, SEQ ID NO:71, SEQ ID NO:7, SEQ ID NO:8, SEQ ID NO:9, SEQ ID NO:10, SEQ ID NO:11, SEQ ID NO:12, SEQ ID NO:13, SEQ ID NO:14, SEQ ID NO:15, SEQ ID NO:16, SEQ ID NO:17, SEQ ID NO:18, SEQ ID NO:19, SEQ ID NO:20, SEQ ID NO:21, and biologically active analogs thereof.

53. The isolated polynucleotide of claim 52 wherein said polynucleotide comprises a member selected from the group consisting of the nucleotide sequence as set forth in SEQ ID NO:40, SEQ ID NO:41, SEQ ID NO:72, SEQ ID NO:42, SEQ ID NO:43, SEQ ID NO:44, SEQ ID NO:45, SEQ ID NO:46, SEQ ID NO:47, SEQ ID NO:48, SEQ ID NO:49, SEQ ID NO:50, SEQ ID NO:51, SEQ ID NO:52, SEQ ID NO:53, SEQ ID NO:54, SEQ ID NO:55, SEQ ID NO:56, SEQ ID NO:85, SEQ ID NO:86, SEQ ID NO:87, SEQ ID NO:88, SEQ ID NO:89, SEQ ID NO:90, SEQ ID NO:91 and SEQ ID NO:92.

54. The isolated polynucleotide of claim 52 wherein said polynucleotide is capable of hybridizing to and which is at least about 70% identical to the nucleotide sequence as set forth in SEQ ID NO:40, SEQ ID NO:41, SEQ ID NO:42 or SEQ ID NO:43, SEQ ID NO:44, SEQ ID NO:45, SEQ ID NO:46, SEQ ID NO:47, SEQ ID NO:48, SEQ ID NO:49, SEQ ID NO:50, SEQ ID NO:51, SEQ ID NO:52, SEQ ID NO:53, SEQ ID NO:54, SEQ ID NO:55, SEQ ID NO:56, SEQ ID NO:85, SEQ ID NO:86, SEQ ID NO:87, SEQ ID NO:88, SEQ ID NO:89, SEQ ID NO:90, SEQ ID NO:91 or SEQ ID NO:92.

55. An isolated polynucleotide comprising a member selected from the group consisting of:

a polynucleotide encoding the peptide comprising the amino acid sequence as set forth in SEQ ID NO:22, SEQ ID NO:23, SEQ ID NO:24, SEQ ID NO:25, SEQ ID NO:26, SEQ ID NO:27, SEQ ID NO:28, and biologically active analogs thereof.

56. The isolated polynucleotide of claim 55 wherein said polynucleotide comprises a member selected from the group consisting of the nucleotide sequence as set forth in SEQ ID NO:57, SEQ ID NO:58, SEQ ID NO:59, SEQ ID NO:60, SEQ ID NO:61, SEQ ID NO:62 and SEQ ID NO:63.

57. The isolated polynucleotide of claim 55 wherein said polynucleotide is capable of hybridizing to and which is at least about 70% identical to the nucleotide sequence as set forth in SEQ ID NO:57, SEQ ID NO:58, SEQ ID NO:59, SEQ ID NO:60, SEQ ID NO:61, SEQ ID NO:62 or SEQ ID NO:63.

58. An isolated polynucleotide comprising a member selected from the group consisting of:

a polynucleotide encoding the peptide comprising the amino acid sequence as set forth in SEQ ID NO:29, SEQ ID NO:30, SEQ ID NO:31, SEQ ID NO:32, SEQ ID NO:33, SEQ ID NO:34, SEQ ID NO: 35, and biologically active analogs thereof.

59. The isolated polynucleotide of claim 58 wherein said polynucleotide comprises a member selected from the group consisting of the nucleotide sequence as set forth in SEQ ID NO:64, SEQ ID NO:65, SEQ ID NO:66, SEQ ID NO:67, SEQ ID NO:68, SEQ ID NO:69 and SEQ ID NO:70.

60. The isolated polynucleotide of claim 58 wherein said polynucleotide is capable of hybridizing to and which is at least about 70% identical to the nucleotide sequence as set forth in SEQ ID NO:64, SEQ ID NO:65, SEQ ID NO:66 or SEQ ID NO:67, SEQ ID NO:68, SEQ ID NO:69 or SEQ ID NO:70.

61. A gene therapy vector comprising a first nucleotide sequence encoding a peptide fragment of the conserved regulatory domain of NFAT protein capable of inhibiting protein-protein interaction between calcineurin and NFAT, or a biologically active analog of said peptide fragment.

62. The gene therapy vector of claim 61 wherein said first nucleotide sequence comprises a member selected from the group consisting of:

a nucleotide sequence encoding the peptide comprising the

amino acid sequence as set forth in SEQ ID NO:105, SEQ ID NO:106, SEQ ID NO:107, SEQ ID NO:108, and biologically active analogs thereof.

63. The gene therapy vector of claim 62 wherein said first nucleotide sequence comprises a member selected from the group consisting of:

the nucleotide sequence as set forth in SEQ ID NO:110, SEQ ID NO:111, SEQ ID NO:112, SEQ ID NO:113, SEQ ID NO:114 and SEQ ID NO:115.

64. The gene therapy vector of claim 62 wherein said nucleotide sequence is capable of hybridizing to and which is at least about 70% identical to the nucleotide sequence as set forth in SEQ ID NO:110, SEQ ID NO:111, SEQ ID NO:112, SEQ ID NO:113, SEQ ID NO:114 or SEQ ID NO:115.

65. The gene therapy vector of claim 61 wherein said first nucleotide sequence comprises a member selected from the group consisting of:

a nucleotide sequence encoding the peptide comprising the amino acid sequence as set forth in SEQ ID NO:1, SEQ ID NO:2, SEQ ID NO:3, SEQ ID NO:4, and biologically active analogs thereof.

66. The gene therapy vector of claim 65 wherein said first nucleotide sequence comprises a member selected from the group consisting of:

the nucleotide sequence as set forth in SEQ ID NO:36, SEQ ID NO:37, SEQ ID NO:38, SEQ ID NO:39, SEQ ID NO:83 and SEQ ID NO:84.

67. The gene therapy vector of claim 65 wherein said nucleotide sequence is capable of hybridizing to and which is at least about 70% identical to the nucleotide sequence as set forth in SEQ ID NO:36, SEQ ID NO:37, SEQ ID NO:38, SEQ ID NO:39, SEQ ID NO:83 or SEQ ID NO:84.

68. The gene therapy vector of claim 61 wherein said first nucleotide sequence comprises a member selected from the group consisting of:

a nucleotide sequence encoding the peptide comprising the amino acid sequence as set forth in SEQ ID NO: 5, SEQ ID NO: 6, SEQ ID NO:71, SEQ ID NO:7, SEQ ID NO:8, SEQ ID NO:9, SEQ ID NO:10, SEQ ID NO:11, SEQ ID NO:12, SEQ ID NO:13, SEQ ID NO:14, SEQ ID NO:15, SEQ ID NO:16, SEQ ID NO:17, SEQ ID NO:18, SEQ ID NO:19, SEQ ID NO:20, SEQ ID NO:21, and biologically active analogs thereof.

69. The gene therapy vector of claim 68 wherein said first nucleotide sequence comprises a member selected from the group consisting of:

the nucleotide sequence as set forth in SEQ ID NO:40, SEQ ID NO:41, SEQ ID NO:72, SEQ ID NO:42, SEQ ID NO:43, SEQ ID NO:44, SEQ ID NO:45, SEQ ID NO:46, SEQ ID NO:47, SEQ ID NO:48, SEQ ID NO:49, SEQ ID NO:50, SEQ ID NO:51, SEQ ID NO:52, SEQ ID NO:53, SEQ ID NO:54, SEQ ID NO:55, SEQ ID NO:56, SEQ ID NO:85, SEQ ID NO:86, SEQ ID NO:87, SEQ ID NO:88, SEQ ID NO:89, SEQ ID NO:90, SEQ ID NO:91 or SEQ ID NO:92.

70. The gene therapy vector of claim 68 wherein said nucleotide sequence is capable of hybridizing to and which is at least about 70% identical to the nucleotide sequence as set forth in SEQ ID NO:40, SEQ ID NO:41, SEQ ID NO:42, SEQ ID NO:43, SEQ ID NO:44, SEQ ID NO:45, SEQ ID NO:46, SEQ ID NO:47, SEQ ID NO:48, SEQ ID NO:49, SEQ ID NO:50, SEQ ID NO:51, SEQ ID NO:52, SEQ ID NO:53, SEQ ID NO:54, SEQ ID NO:55, SEQ ID NO:56, SEQ ID NO:85, SEQ ID NO:86, SEQ ID NO:87, SEQ ID NO:88, SEQ ID NO:89, SEQ ID NO:90, SEQ ID NO:91 or SEQ ID NO:92.

71. The gene therapy vector of claim 61 wherein said first nucleotide sequence comprises a member selected from the group consisting of:

a nucleotide sequence encoding the peptide comprising the amino acid sequence as set forth in SEQ ID NO:22, SEQ ID NO:23, SEQ ID NO:24, SEQ ID NO:25, SEQ ID NO:26, SEQ ID NO:27, SEQ ID NO:28, and biologically active analogs thereof.

72. The gene therapy vector of claim 71 wherein said first nucleotide sequence comprises a member selected from the group consisting of:

the nucleotide sequence as set forth in SEQ ID NO:57, SEQ ID NO:58, SEQ ID NO:59, SEQ ID NO:60, SEQ ID NO:61, SEQ ID NO:62 and SEQ ID NO:63.

73. The gene therapy vector of claim 71 wherein said nucleotide sequence is capable of hybridizing to and which is at least about 70% identical to the nucleotide sequence as set forth in SEQ ID NO:57, SEQ ID NO:58, SEQ ID NO:59, SEQ ID NO:60, SEQ ID NO:61, SEQ ID NO:62 or SEQ ID NO:63.

74. The gene therapy vector of claim 61 wherein said first nucleotide sequence comprises a member selected from the group consisting of:

a nucleotide sequence encoding the peptide comprising the amino acid sequence as set forth in SEQ ID NO:29, SEQ ID NO:30, SEQ ID NO:31, SEQ ID NO:32, SEQ ID NO:33, SEQ ID NO:34 and SEQ ID NO:35, and biologically active analogs thereof.

75. The gene therapy vector of claim 74 wherein said first nucleotide sequence comprises a member selected from the group consisting of:

the nucleotide sequence as set forth in SEQ ID NO:64, SEQ ID NO:65, SEQ ID NO:66, SEQ ID NO:67, SEQ ID NO:68, SEQ ID NO:69 and SEQ ID NO:70.

76. The gene therapy vector of claim 74 wherein said nucleotide sequence is capable of hybridizing to and which is at least about 70% identical to the nucleotide sequence as set forth in SEQ ID NO:64, SEQ ID NO:65, SEQ ID NO:66, SEQ ID NO:67, SEQ ID NO:68, SEQ ID NO:69 or SEQ ID NO:70.

77. The gene therapy vector of claim 61 further comprising a second nucleotide sequence encoding a signal peptide for effecting secretion of said peptide fragment.

78. The gene therapy vector of claim 61 further comprising a third nucleotide sequence encoding a tag for identification of said peptide fragment.

79. The gene therapy vector of claim 61 further comprising a selectable marker.

80. A cell comprising said gene therapy vector of claim 61.

81. A method for producing a peptide capable of inhibiting protein-protein interaction between calcineurin and NFAT, comprising culturing a cell of claim 80 under conditions that permit expression of said peptide.

82. A method for treating an immune-related disease or condition in an animal, comprising administering to said animal said gene therapy vector of claim 61.

83. A method for providing an animal having an immune-related disease or condition with a therapeutically effective level of a peptide capable of inhibiting protein-protein interaction between calcineurin and NFAT, comprising administering to said animal said gene therapy vector of claim 61.

84. A method for inhibiting an immune response in an animal, comprising;

providing an animal in need of inhibition of an immune response;

providing a therapeutically effective amount of a peptide fragment of the conserved regulatory domain of NFAT protein capable of inhibiting protein-protein interaction between calcineurin and NFAT, or a biologically active analog thereof;

and

administering said peptide fragment or said biologically active analog thereof to said animal so as to inhibit said immune response in said animal.

85. The method of claim 84 wherein said peptide fragment is further capable of inhibiting dephosphorylation of said NFAT by said calcineurin.

86. The method of claim 84 wherein said NFAT protein is selected from the group consisting of NFAT1, NFAT2, NFAT3 and NFAT4.

87. The method of claim 84 wherein said peptide fragment comprises the amino acid sequence IX_2X_3T (SEQ ID NO:104) or a biologically active analog thereof, wherein X_2 is E, R or Q, and X_3 is I or F.

88. The method of claim 84 wherein said peptide fragment comprises the amino acid sequence selected from the group consisting of SEQ ID NO:105, SEQ ID NO:106, SEQ ID NO:107, SEQ ID NO:108, and biologically active analogs thereof.

89. The method of claim 84 wherein said peptide fragment comprises the amino acid sequence $X_1IX_2X_3T$ (SEQ ID NO:73) or a biologically active analog thereof, and wherein X_1 is R or S; X_2 is E, R or Q, and X_3 is I or F.

90. The method of claim 84 wherein said peptide fragment comprises the amino acid sequence selected from the group consisting of SEQ ID NO:1, SEQ ID NO:2, SEQ ID NO:3, SEQ ID NO:4, and biologically active analogs thereof.

91. The method of claim 84 wherein said therapeutically effective amount of said peptide fragment is provided by providing to said animal a recombinant nucleic acid having a nucleotide sequence encoding said peptide fragment or biologically active analog thereof and which is capable of expressing said peptide fragment or biologically active analog thereof in vivo.

92. The method of claim 91 wherein said recombinant nucleic acid is a gene therapy vector.

93. The method of claim 84 wherein said therapeutically effective amount of said peptide fragment is provided by providing to said animal a composition comprising animal cells wherein a recombinant nucleic acid having a nucleotide sequence encoding said peptide fragment has been introduced ex vivo into said animal cells so as to express said peptide fragment in said animal cells.

94. The method of claim 93 wherein said recombinant nucleic acid is a gene therapy vector.

95. The method of claim 93 wherein said animal cells are selected from the group consisting of cells derived from said animal and allogeneic cells.

96. A method for treating a disease involving hyperactivity or inappropriate activity of the immune system, a transplant graft rejection or graft-versus-host disease in an animal, comprising:

providing an animal in need of treatment for a disease involving hyperactivity or inappropriate activity of the immune system, a transplant graft rejection or graft-versus-host disease;

providing a therapeutically effective amount of a peptide fragment of the conserved regulatory domain of NFAT protein capable of inhibiting protein-protein interaction between calcineurin and NFAT, or a biologically active analog thereof; and

administering said peptide fragment or biologically active analog thereof to said animal in a therapeutically effective amount such that treatment of said disease involving hyperactivity or inappropriate activity of the immune system, said transplant graft rejection or said graft-versus-host disease occurs.

97. The method of claim 96 wherein said disease involving hyperactivity or inappropriate activity of the immune system is selected from the group consisting of an acute immune disease, a chronic immune disease and an autoimmune disease.

98. The method of claim 96 wherein said therapeutically effective amount of said peptide fragment or biologically active analog thereof is administered by providing to said animal a nucleic acid encoding said peptide fragment or biologically

active analog thereof and expressing said peptide fragment or biologically active analog thereof in vivo.

99. The method of claim 96 wherein said peptide fragment comprises the amino acid sequence IX_2X_3T (SEQ ID NO:104) or a biologically active analog thereof, and wherein X_2 is E, R or Q, and X_3 is I or F.

100. The method of claim 96 wherein said peptide fragment comprises the amino acid sequence selected from the group consisting of SEQ ID NO:105, SEQ ID NO:106, SEQ ID NO:107, SEQ ID NO:108, and biologically active analogs thereof.

101. The method of claim 96 wherein said peptide fragment comprises the amino acid sequence $X_1IX_2X_3T$ (SEQ ID NO:73) or a biologically active analog thereof, and wherein X_1 is R or S; X_2 is E, R or Q, and X_3 is I or F.

102. The method of claim 96 wherein said peptide fragment comprises the amino acid sequence selected from the group consisting of SEQ ID NO:1, SEQ ID NO:2, SEQ ID NO:3, SEQ ID NO:4, and biologically active analogs thereof.

103. A method for treating an animal at risk for a disease involving hyperactivity or inappropriate activity of the immune system, a transplant graft rejection or graft-versus-host disease in an animal, comprising:

providing an animal at risk for a disease involving hyperactivity or inappropriate activity of the immune system, a

transplant graft rejection or graft-versus-host disease;

providing a therapeutically effective amount of a peptide fragment of the conserved regulatory domain of NFAT protein capable of inhibiting protein-protein interaction between calcineurin and NFAT, or a biologically active analog thereof; and

administering said peptide fragment or biologically active analog thereof to said animal in a therapeutically effective amount such that treatment occurs.

104. A method for gene therapy comprising genetically modifying an animal cell such that it is able to express a peptide fragment or biologically active analog thereof of the conserved regulatory domain of NFAT protein, said peptide fragment being capable of inhibiting calcineurin-mediated NFAT activation, so as to effect gene therapy.

105. The method of claim 104 wherein said animal cells are genetically modified by introducing into said cells a recombinant nucleic acid having a nucleotide sequence encoding said peptide fragment and which is capable of expressing said peptide fragment in vivo.

106. A pharmaceutical composition for treating an immune-related disease or condition in an animal, comprising:

a therapeutically effective amount of a peptide fragment of the conserved regulatory domain of NFAT protein capable of inhibiting protein-protein interaction between calcineurin and NFAT, or a biologically active analog thereof; and

a pharmaceutically acceptable carrier.

107. A pharmaceutical composition for treating an immune-related disease or condition in an animal, comprising:

a therapeutically effective amount of a recombinant nucleic acid having a nucleotide sequence encoding a peptide fragment of the conserved regulatory domain of NFAT protein capable of inhibiting protein-protein interaction between calcineurin and NFAT, or a biologically active analog thereof; and

a pharmaceutically acceptable carrier.

108. A pharmaceutical composition for treating an immune-related disease or condition in an animal, comprising:

a therapeutically effective amount of animal cells wherein a recombinant nucleic acid having a nucleotide sequence encoding a peptide fragment of the conserved regulatory domain of NFAT protein capable of inhibiting protein-protein interaction between calcineurin and NFAT, or a biologically active analog thereof, has been introduced into said animal cells so as to express said peptide fragment; and

a pharmaceutically acceptable carrier.

109. The pharmaceutical composition of claim 108 wherein said animal cells are selected from the group consisting of cells derived from said animal to be treated and allogeneic cells.

110. A method for inhibiting protein-protein interaction between calcineurin and NFAT <u>in vivo</u>, comprising:

providing a cell having calcineurin and NFAT;

providing a peptide fragment or a biologically active analog thereof of the conserved regulatory domain of NFAT protein

capable of inhibiting protein-protein interaction between calcineurin and NFAT; and

contacting said calcineurin and said peptide fragment or said biologically active analog thereof in vivo such that protein-protein interaction between said calcineurin and said NFAT is inhibited.

111. A method for inhibiting protein-protein interaction between calcineurin and NFAT in vitro, comprising:

providing calcineurin and NFAT;

providing a peptide fragment or a biologically active analog thereof of the conserved regulatory domain of NFAT protein capable of inhibiting protein-protein interaction between calcineurin and NFAT; and

contacting said calcineurin and said peptide fragment or said biologically active analog thereof in vitro such that protein-protein interaction between said calcineurin and said NFAT is inhibited.

112. A method for evaluating an agent for use in modulating an immune response, comprising:

providing a cell;

providing an agent selected from the group consisting of a peptide fragment of the conserved regulatory domain of NFAT protein and biologically active analogs thereof; and

evaluating the effect of said agent on an aspect of calcineurin-mediated NFAT activation, a change in said aspect of calcineurin-mediated NFAT activation being indicative of the usefulness of said agent in modulating an immune response.

113. A method for high-throughput screening of candidate agents to identify an agent that inhibits protein-protein interaction between calcineurin and NFAT, comprising:

providing a first compound selected from the group consisting of calcineurin or a biologically active derivative thereof, and NFAT or a biologically active derivative thereof;

providing a second compound selected from the group consisting of calcineurin or a biologically active derivative thereof, and NFAT or a biologically active derivative thereof, wherein said second compound is different from said first compound, and wherein said second compound is labeled;

providing a candidate agent;

contacting said first compound, said second compound and said agent with each other; and

determining the amount of label bound to said first compound, wherein a reduction in protein-protein interaction between said first compound and said second compound as assessed by label bound is indicative of the usefulness of said agent in inhibiting protein-protein intereaction between calcineurin and NFAT.

114. The method of claim 113 wherein said first compound that is provided is attached to a solid support.

115. The method of claim 113 further comprising a washing step after said contacting step, so as to separate bound and unbound label.

116. The method of claim 113 wherein said second compound is labeled with a radiolabel.

117. The method of claim 113 wherein a plurality of candidate agents are contacted with said first compound and said second compound.

118. The method of claim 117 wherein each of said candidate agents is contacted with said first compound and said second compound in separate wells.

119. The method of claim 113 wherein said first compound is calcineurin or a biologically active derivative thereof, and said second compound is NFAT or a biologically active derivative thereof.

120. The method of claim 113 wherein said first compound is NFAT or a biologically active derivative thereof, and said second compound is calcineurin or a biologically active derivative thereof.

121. The method of claim 113 wherein said NFAT derivative is a mutated NFAT having increased affinity for calcineurin.

122. The method of claim 113 wherein said NFAT derivative is a peptide fragment of the conserved regulatory domain of NFAT protein capable of interacting with calcineurin.

123. A method for high-throughput screening of candidate agents to identify an agent that inhibits dephosphorylation of NFAT by calcineurin, comprising:

providing phosphorylated NFAT;

providing calcineurin or a biologically active derivative thereof having enzymatic activity;

providing a candidate agent;

contacting said phosphorylated NFAT, said calcineurin or biologically active derivative thereof, and said candidate agent, with each other in reaction media under conditions that allow enzymatic activity of calcineurin; and

determining whether phosphate remains associated with said NFAT, wherein if phosphate remains associated with said NFAT it is indicative of the usefulness of said agent in inhibiting dephosphorylation of NFAT by calcineurin.

124. The method of claim 123 further comprising the step of separating said NFAT from said reaction media after the contacting step.

125. The method of claim 123 wherein said phosphorylated NFAT that is provided is attached to a solid support.

126. The method of claim 123 wherein a plurality of candidate agents are contacted with said phosphorylated NFAT and said calcineurin or biologically active derivative thereof.

127. The method of claim 126 wherein each of said candidate agents is contacted with said phosphorylated NFAT and said

calcineurin or biologically active derivative thereof, in separate wells.

128. The method of claim 123 wherein said phosphorylated NFAT that is provided is labeled phosphorylated NFAT.

129. The method of claim 128 wherein said labeled phosphorylated NFAT is radiolabeled phosphorylated NFAT.

130. The method of claim 128 wherein the determining step comprises determining the release of labeled phosphate in said reaction media or the retention of labeled phosphate on said NFAT, wherein a reduction in release of labeled phosphate from said NFAT by calcineurin, or an increase in retention of labeled phosphate on said NFAT, is indicative of the usefulness of said agent in inhibiting dephosphorylation of NFAT by calcineurin.

131. The method of claim 123 further comprising the step of determining inhibition of dephosphorylation by said agent of a calcineurin substrate other than NFAT by calcineurin.

132. The method of claim 123 wherein the determining step comprises using antibodies or a functionally equivalent reagent that discriminates between phosphorylated and unphosphorylated forms of NFAT peptides.

133. A method for high-throughput screening of candidate agents to identify an agent that inhibits conformational change in NFAT from dephosphorylation by calcineurin, comprising:

providing phosphorylated NFAT;

providing calcineurin or a biologically active derivative thereof having enzymatic activity;

providing a candidate agent;

providing an oligonucleotide having an NFAT site;

contacting said phosphorylated NFAT, said calcineurin or biologically active derivative thereof, and said candidate agent with each other in reaction media under conditions that allow enzymatic activity of calcineurin; and

determining specific binding of said NFAT to said oligonucleotide having said NFAT site, wherein a reduction of binding is indicative of the usefulness of said agent in inhibiting conformational change in NFAT from dephosphorylation by calcineurin.

134. The method of claim 133 wherein said phosphorylated NFAT that is provided is attached to a solid support.

135. A method for high-throughput screening of candidate agents to identify an agent that inhibits calcineurin-dependent import of NFAT into the nucleus of a cell, comprising:

providing cells expressing NFAT;

providing a stimulant that activates NFAT through the calcium/calcineurin pathway;

providing a candidate agent:

contacting said cells, said stimulant and said candidate agent with each other; and

determining the presence or absence of nuclear NFAT in said cells, wherein a reduction in nuclear NFAT is indicative of the

agent inhibiting calcineurin-dependent import of NFAT into the nucleus of a cell.

136. A method for assessing the state of NFAT activation of immune system cells isolated from an animal, comprising:

providing immune system cells isolated from an animal; and

determining the presence or absence of nuclear NFAT in said cells, wherein the presence of nuclear NFAT in said cells is indicative of activation of NFAT in said cells.

137. The method of claim 136 wherein said cells are isolated by biopsy or aspiration of said cells from said animal.

138. The method of claim 136 wherein the presence or absence of nuclear NFAT in said cells is determined by histological staining of said cells.

139. The method of claim 136 wherein said cells are infiltrating cells at a site of inflammation or in a tumor.

140. A method for assessing the ability of immune system cells isolated from an animal to respond to an NFAT activating signal, comprising:

providing immune system cells from an animal, said cells being unactivated for NFAT;

providing a stimulant that activates NFAT;

contacting said cells with said stimulant; and

determining the presence or absence of nuclear NFAT in said cells, wherein a reduction in nuclear NFAT is indicative of

impairment of the ability of said cells to respond to an NFAT activating signal.

141. The method of claim 140 wherein said animal is immunocompromised.

142. A method for identifying a stimulant that can activate NFAT in immune system cells isolated from an animal, comprising:

providing immune system cells isolated from an animal;

providing a candidate stimulant;

contacting said cells with said stimulant; and

determining the presence or absence of nuclear NFAT in said cells, wherein the presence of nuclear NFAT is indicative of said stimulant activating NFAT in said cells.

143. The method of claim 142 wherein said stimulant is an allergen.

144. A method for identifying an allergen, comprising:

providing an animal cell line expressing NFAT;

providing IgE from an animal;

providing a candidate allergen;

contacting said cell line with said IgE;

contacting said cell line with said candidate allergen; and

determining the presence or absence of nuclear NFAT in cells of said cell line, wherein the presence of nuclear NFAT is indicative of said candidate allergen being an allergen.